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GENOTYPIC DIVERSITY OF POTATO GERmplasm AGAINST ROOT KNOT NEMATODE (*MELOIDOGYNE INCOGNITA*) INFECTION IN PAKISTAN

Amjad S. Gondal*, Nazir Javed, Sajid A. Khan, Sajjad Hyder

Department of Plant Pathology, University of Agriculture Faisalabad, Pakistan.

*Corresponding Author Email: amjadshahzad@live.com

ABSTRACT

Potato (*Solanum tuberosum*), an important vegetable crop of Pakistan endures significant yield losses due to root knot nematode (*Meloidogyne incognita*). Research work was designed to identify resistant potato germplasm against RKN (*Meloidogyne incognita*) infection. A field trial was conducted in the research area of Department of Plant Pathology, University of Agriculture Faisalabad. Thirty six (36) potato varieties/ cultivars relocated five times were sown in four years sick plot containing root knot nematode (*Meloidogyne incognita*) in RCBD layout. Root knot nematode reproduction and host damage was assessed by recording nematode root galls and egg mass indices, root weight, shoot weight, , number of leaves, fruit weight, rate of reproduction and final population of nematodes. Experiment revealed a considerable variation in response against *Meloidogyne incognita* infection among the genotype tested but none of the single cultivar was immune. FD-8-1 was used as negative control. The cultivar FD-19-2 was highly susceptible followed by SH-692 and SH-5. All other cultivars had less galling index with low fecundity rate indicating their ability to suppress the adult female reproduction. The cultivar FD-1-3 scored least number of galls and egg mass indices followed by FD-49-62, SH-339 and SH-332.

Keywords: Root knot nematode, *Meloidogyne incognita*, Genotypic response, Potato, *Solanum tuberosum*.

INTRODUCTION

Potato (*Solanum tuberosum* L.) is perennial solanaceous vegetable crop also known as nightshades. It is grown in 140 countries of the world (Haase, 2008). It endures significant yield losses due to biotic and abiotic stresses (Agrios, 2005). It is attacked by numerous fungal, bacterial, viral and nematode diseases that reduce both quality and quantity of the produce. Among biological agents, nematodes especially root knot nematodes (RKN) inflict heavy yield reduction (Anwar *et al.*, 2009). Root knot nematodes are parasitic on a broad range of vegetable crops. They stand out as the major group of plant parasitic nematodes in almost all vegetable crops especially on potato and cause enormous yield losses (Mehrotra, 1983). Root knot nematodes (*Meloidogyne incognita*) are endoparasites that are distributed in tropical, subtropical and warmer regions of the world where the climate is suitable for their reproduction throughout the year. *Meloidogyne* spp. with an approximate distribution of *M. incognita* 58%, *M.*

javanica 31%, *M. aranaria* 8%, *M. hapla* 7% and other species are about 2% have been recorded in the agricultural soils of Pakistan (Maqbool, 1986). The presence of root knot nematodes with different vegetables, ornamental & medicinal plants and fruit trees have been reported in Pakistan and other countries (Das and Das, 2000; Ravichandra and Krishnappa, 2004; Khan *et al.*, 2006; Arooj, 2011). Two species of root knot nematodes; *M. incognita* and *M. javanica* have been reported to parasitize potato in almost all potato growing areas of Punjab (Anwar *et al.*, 1991; Khan and Ahmad, 2000; Anwar *et al.*, 2007).

M. incognita cause more losses to the vegetables especially to potato crops grown in the sandy loam soils of Punjab (Anwar and Mckenry, 2006). Root knot nematodes are amongst the damaging of plant parasitic nematodes. The genus *Meloidogyne* has more than 80 known species (Karseen, 2000). The most destructive species in the tropics and the subtropics include *M. javanica*, *M. aranaria* and *M. incognita* (Sikora and

Fernandez, 2005). On account of great losses, they require immediate and due attention to control its predation (Rehman *et al.*, 2006).

Root knots and gall formation of variable size is the characteristic symptoms of root knot nematode infection. Root galls cause disturbance in nutrient absorption, water essential elements uptake from the soil that leads to the poor plant growth ultimately severe yield losses (Hollis, 1963; Gowen *et al.*, 2005). up to 95% annual yield losses were observed in vegetables due to high infestation of *Meloidogyne incognita* spp. (Bourne *et al.*, 2004; Cetintas and Yarba, 2010).

Root knot nematodes are problematic to control because of their high population densities and reproductive potential (Sikora and Fernandez, 2005). RKN population was determined from different vegetable crops including tomato, potato, cucumber and squash (Karajeh *et al.*, 2005; Kepenekci and Evlice, 2004).

An efficient and environment friendly way to prevent widespread destruction by nematode infections is to incorporate genotypic natural resistance in potato germplasm. The use of resistant genotypes is an attractive alternate for managing yield losses caused by plant parasitic nematodes which does not require major adaptations in the farming practices (Younis *et al.*, 2009).

Resistant plant inhibits the reproduction of nematode species. Resistance of plant to nematode may be defined as plant ability to inhibit the attack of nematode. Resistance is widely and effectively used in vegetable and field crops (Anwar *et al.*, 2007) such as tomato (Khan *et al.*, 2000; Amin, 2003; Sajjad, 2004; Anwar *et al.*, 2007; Abbas *et al.*, 2008), Sugar beet (Zia *et al.*, 2008), Chilli (Anwar *et al.*, 2007) Cucumber (Anwar *et al.*, 2007) and Soybean (Anwar, 1996; Ashiq *et al.*, 2004). Different germplasm responds differently to diseases and pest attacks. The variation in response to nematode infection in different cultivars could be related to their genetic makeup and the level of resistance mechanism possessed by a particular cultivar (Abad *et al.*, 2003; Barham and Winstead, 1957; Anwar and Mckenry, 2002). Varietal response against *M. incognita* infection of different vegetables is accessed on the basis of plant growth parameters including root weight, shoot weight, shoot height, number of leaves per plant and nematode reproduction parameters including number of galls, egg masses, number of females, rate of reproduction and final population of nematodes (Anwar *et al.*, 2003; Hayat, 2011).

Keeping in view the above mentioned facts, the present study aimed at exploring the source of genotypic resistance among potato germplasm against root knot nematode (*Meloidogyne incognita*).

MATERIALS AND METHODS

Collection of Diseased Samples: Roots of diseased potato plants with typical symptoms of root knot nematode were collected from different potato growing areas of the Punjab province of Pakistan. Samples were collected with the help of trowel 15-20 cm deep from the rhizosphere of the diseased plants together with approximately 1kg of adhering soil. Samples were kept in the polythene bags and moistened well to ensure the adequate moisture for nematode survival. Root samples were stored at 5°C (40°F) in refrigerator in Plant Nematology lab. Department of Plant Pathology, University of Agriculture Faisalabad.

Isolation of Root Knot Nematodes from Roots and Soil: White-head and Hemming tray method (White-head and Hemming, 1965) was used for isolation of *M. incognita* second stage juveniles (J2) from root samples. The entire root system was cut into small pieces, chopped and composite sample of 20 g was placed in the tray lined with tissue paper having sufficient amount of water. After 24 hours water was poured off in a beaker and allowed to settle for one hour. Juveniles were settled down and excess water was drained off remaining the juvenile suspension.

Soil sample was thoroughly mixed and 100cm³ composite samples was placed in plastic bucket. 1 liter of tap water was added and thoroughly mixed to break the clods. Heavy soil particles, roots and rocks were drained off from the bucket with hand. Supernatant water was poured through the coarse sieve (50-mesh) into the second bucket. A liter of water was added and the above step was repeated to get optimum number of nematodes. The water suspension was stirred and allowed to settle down. The supernatant water was then poured through fine sieve (100-mesh) to another bucket. Process was repeated using fine sieves (250 and 325-mesh). Suspension left after passing through 325-mesh sieve was transferred to 500 ml beaker. Suspension was allowed to settle down to bottom of the beaker and access waster siphoned out suing siphon tube.

Identification of root knot nematodes: Root Knot Nematode species were identified on the basis of perineal pattern (Jepson, 1987). Visible knots on the root tissues were dissected longitudinally using sterile

blade to detach female nematode. It was then transferred to glass slide to cut posterior half of its body. Portion containing perineal pattern was obtained, by cutting lower cuticle in a square shape, and transferred to another glass slide containing few drops of glycerin. Paraffin was used to seal the cover slip on the slide. The slide was placed under stereoscopic microscope to observe female perineal pattern to identify *Meloidogyne* species (Eisenback *et al.*, 1981).

Purification of *Meloidogyne incognita* using single egg mass culture: From galled potato roots, egg masses of the uniform size were collected using fine forceps. These egg masses were stored in petri dishes in refrigerator at 4°C. One month old potato seedlings were inoculated with these egg masses @ one egg mass per plant near the root zone making a hole with the help of pointed wood avoiding the root injury. The hole was covered with soil to avoid drying. Plants were watered regularly to prevent loss of nematodes through drying or leaching.

Rearing of Root Knot Nematode (*M. incognita*): Mass culturing of *M. incognita* was done on the roots of the susceptible potato cultivar FD-8-1 in order to get regular supply of inoculum for experiments. Potato plants were planted in 20 cm diameter earthen pots containing formalin sterilized sandy loam soil. Approximate of 2000 eggs of *M. incognita* in a small volume of egg suspension were pipetted around each plant by making 3-4 holes with the help of pointed wood. The holes were then covered with soil to avoid drying. Plants were watered regularly to prevent excess drying.

Soil Preparation: Sandy loam soil containing 70% sand, 22% silt and 8% clay was thoroughly mixed and air dried by speeding in a thin layer on woody bench, covered with plastic sheet. Large stones and plant debris were removed followed by mechanical analysis of the soil.

Soil Sterilization: Soil sterilization was accomplished by the application of formalin. Formalin diluted to 1:320 ratio was added to small heap of soil. The soil was then covered with polythene sheet to stop the fumes completely. After 7 days, the soil was exposed to get rid of residual formalin. The soil was checked to ensure that there was no nematode population after sterilization. Thoroughly mixed soil was filled in the medium sized pots.

Collection of Potato Tubers: Potato seed tubers of different cultivars were collected from Vegetable Research Institute, Faisalabad and Potato Research

Institute, Sahiwal. Care was made for seed health, purity and germinating capacity.

Location of Experiment: The experiment was conducted in the research area of Department of Plant Pathology, University of Agriculture Faisalabad, Pakistan (31°25' 0" N/ 73° 5' 0" E).

Evaluation Experiments

Counting and Standardization of Eggs: To estimate the inoculum density, nematode suspension was poured into measuring cylinder. The suspension was mixed vigorously by blowing and stirring with pipette. The numbers of nematodes were estimated in 1-ml aliquots in a counting slide under binomial stereoscopic microscope. The process was repeated three times and total population was estimated by obtaining means of the three aliquots.

Estimation of Nematode Numbers in Roots: Estimation of nematode numbers in roots was based on whole root systems. It is considered as a general practice in nematology to take (mixed) root samples for estimating the number of nematodes. However, in our study only small potted-plants, inoculated with relatively small nematode numbers were used and counting the whole sample was regarded as most accurate and reliable. Two methods were used; staining nematodes within the root system and extracting nematodes from the root system.

Estimation of nematode suspensions: When counting and estimating nematode suspensions, all samples were made up to equal volumes for a given experiment. Volumes varied from 25 to 300 ml. Samples too large in volume were left undisturbed for several hours, allowing nematodes to settle on the bottom. Water was then siphoned off until the volume was reduced to the required level. Before taking each aliquot, the suspension was agitated to ensure an even distribution of nematodes. For each suspension, several 1, 2 or 3 ml aliquots were taken and placed in 5.5 cm x 4 cm counting dishes with 2 mm x 4 mm squares. For macerated root suspensions, a pipette with a wide (2 mm) opening was used. The number of nematodes in each aliquot was counted using stereomicroscope (Olympus SZ 61 at 6X magnifications). Estimation could thus be made of the total number of nematodes in the suspension.

Extraction method: Both Whitehead and Hemming tray and Modified Baermann trays methods were used for the extraction of J2 from root material (Whitehead and Hemming, 1965; Southey, 1986). These extraction

methods are based on nematode motility. Modified Baermann extraction trays consisted of sieves made out of nylon gauze fixed onto a plastic ring (11 cm diameter, 3 cm deep). A layer of tissue paper was placed on top of the nylon gauze onto which the root system (cut up into 1-2 cm pieces) or other material was placed. Sieves with 1.5 cm supports fixed onto them were placed in 17 cm diameter, 3 cm deep trays. Trays were then filled thirty six with water until material on the sieves were almost awash. Each extraction tray was covered and incubated at 25-28 °C. Water was carefully added to trays when levels were low. For extraction of nematodes from root material, trays were left for 1-2 weeks and harvested for nematodes two to three times during this period. By removing the sieve from the tray and pouring off the suspension into a beaker, nematodes that migrated out of the root pieces into the water were collected. After collecting nematodes, trays were refilled with water ready for the next harvest.

Staining Methods: Acid Fuchsin and Phloxin B staining methods were used. Acid Fuchsin was used for staining nematodes within root systems and Phloxin B was used to stain *M. incognita* egg masses.

Extraction of *M. incognita* from Infected Seedlings: Infected roots with egg masses were washed out two weeks in advance. Plant roots with 56-63 days old egg masses were obtained in order to get maximum number of mature eggs. Roots were cut into 2-3 cm segments and shaken well for 3-4 minutes in 1L beaker having 200 ml of 1% sodium hypochlorite (NaOCl) solution to dissolve the gelatinous matrix and to release eggs from the egg masses (Adegbite and Adesiyun, 2005). The suspension was poured to a series of sieves to separate the organic debris from eggs. This suspension was quickly passed through 200-mesh (75 µm) sieve nested over 500-mesh (25 µm) sieve to collect root fragments on the former and freed eggs on the latter. The eggs collected on the 500-mesh sieve were rinsed with tap water to remove the residual NaOCl. Rinsing of eggs was done for several minutes. These freed eggs were collected in a 500 ml beaker. This process was repeated twice to get additional eggs and to get rid of residual Chlorax and then incubated at 28 °C. The hatched J2 were collected daily in 500ml beakers and labeled the date of harvesting of eggs. The age of second stage juveniles was thus known. Only freshly hatched J2 collected within 48 h were used for experiments (Hussey and Barker, 1973; Radewald *et al.*, 2003).

Varietal Response of Potato Cultivars against Root

Knot Nematode: Seeds of different potato cultivars were planted at 70 cm between row spacing and 30 cm within row spacing in four years sick plot containing root knot nematode (*M. incognita*). Each treatment was replicated five times. Plants were allowed to grow for 3 months. Irrigation was done weekly throughout the period of research. Growth of the plants was observed regularly. After 90 days, plants were harvested from the field. Roots and shoots of the plants were cut, gently washed in tap water, dump-dried and weighed. Care was taken in order to limit the loss of small roots and egg masses during the washing procedure. To facilitate counting of egg masses, the washed roots were stained with phloxine B (Al-Hazmi and Sasser, 1982; Holbrook *et al.*, 1983; Southey, 1986). Roots were placed in a phloxine B solution (0.15g/ liter tap water) for about 15-20 minutes. The stain was absorbed by the gelatinous matrix, which took a pink to red color while roots remained either unstained or very lightly stained, whereas eggs remained viable. Excess stain was removed by three consecutive rinses in one liter beaker filled with water. After staining, roots were wrapped in tissue paper to prevent drying out during the steps of the procedure of evaluation. Stained egg masses from entire root system were counted under a stereomicroscope (Olympus SZ 61) at 2.5X magnification. The whole root system was rated for galling and egg mass presence on a 0 to 5 scale (Taylor and Sasser, 1978; Quesenberry *et al.*, 1989; Anwar *et al.*, 2007) where 0 = no gall or egg masses, 1 = 1-2, 2 = 3-10, 3 = 11-30, 4 = 31-100, and 5 = >100 galls or egg masses per root system.

The data of following parameters was recorded.

- Fresh weight of root
- Fresh weight of shoot
- Number of galls/root system
- Nematode reproduction factor [Pf/Pi, where Pf = final nematode population at harvest, Pi = initial inoculum].
- Galling index
- Number of egg masses

Grouping of varieties according to level of their resistance or susceptibility was done.

Statistical Analysis: Complete randomized design was used in controlled laboratory conditions and Randomized Complete Block Design was used in the field conditions. SAS Statistical software was used for

the analysis of variance and computing probability (less than 0.05) of the recorded data. Duncan's multiple range tests was used to determine the most significant treatment (Steel *et al.*, 1997).

RESULTS AND DISCUSSION

All thirty seven potato cultivars varied widely in their susceptibility to *M. incognita* infection ($P < 0.05$). This experiment was aimed at to evaluate the genotypic response of thirty six potato cultivars against *M. incognita* as a resistance source for root knot nematode management. Comparison of all these cultivars was accessed on the basis of plant growth parameters (number of leaves per plant, average weight of each tuber, root weight per plant and shoot weight per plant) and nematode reproduction parameters (root galls and egg mass indices, number of nematodes per root system, number nematodes/ 100ml of soil and nematode reproduction factor).

Comparison among different potato cultivars on the basis of plant growth parameters

Effect of *Meloidogyne incognita* infection on number of leaves:

Each potato cultivar responded differently to root knot nematode (*Meloidogyne incognita*) infection. There is a significant difference between numbers of leaves of each plant. Cultivar FD-49-62 had statistically ($P < 0.05$) more number of leaves (75.2) as compared to all other cultivars. FD-1-3, FD-35-25 and FD-53-1 had statistically same number of leaves. Minimum numbers of leaves (29.2) were found in Cardinal followed by FSD-White (30.4) and N-9619 (30.4).

Effect of *Meloidogyne incognita* infection on average tuber weight:

Average tuber weight of each potato cultivar was significantly ($P < 0.05$) different. FD-49-62 had statistically high tuber weight (90.6 g) as compared all other tested cultivars. FD-1-3 and SSH-339 had statistically same tuber weight (62.3 and 61.6 g respectively) higher than that of SH-332 (46.6 g) and FD-51-5 (45.0 g). All other cultivars had statistically low tuber weight due to *M. incognita* infection. Minimum tuber weight was recorded in FD-19-2 (12.0 g) followed by SH-692, FD-8-1 (17.3 g) and SH-5 (23.3 g).

Effect of *Meloidogyne incognita* infection on root weight: FD-49-62 had significantly ($P < 0.05$) less root weight (7.973 g) followed by SH-339, SH-332 and FD-1-3 who had statistically same root weight (8.423, 8.473 and 8.523 g respectively). All other potato cultivars tested against root knot nematode (*Meloidogyne incognita*) infection had statistically more root weight.

Maximum root weight was recorded in FD-8-1 (13.813 g) followed by FD-19-2 (13.783 g) and SH-692 (13.443 g). FD-21-10 (12.873 g) and SH-5 (12.843 g) had statistically same root weight higher than FD-69-1 (12.683 g), SH-216 (12.543 g) and lower than SH-5 potato cultivar.

Effect of *Meloidogyne incognita* infection on shoot weight: Statistically ($P > 0.05$) maximum shoot weight (29.87 g) was recorded in FD-8-3 followed by FD-19-2 (29.46 g), Cardinal (28.13 g), N-22 (27.74 g) and SH-5 (26.96 g). All other cultivars had statistically low shoot weight as compared to these cultivars. Minimum shoot weight (17.9 g) was recorded in FD-49-28. FD-35-1 and SH-339 had statistically same shoot weight (18.61 and 18.60 g respectively).

Comparison among different potato cultivars on the basis of nematode reproduction parameters

Effect of *Meloidogyne incognita* infection on root galls development:

Root knot nematode (*M. incognita*) produced galls on the roots of all thirty six cultivars. Data regarding root galls/root system as shown in Table 2. revealed that statistically ($P < 0.05$) maximum number of galls (44.0) were found on FD-19-2 followed by FD-8-1 (43.33), SH-692 (43.0) and SH-5 (41.33). Statistically same numbers of galls (39.33) were produced on FD-35-25 FD-52-2 and FD-35-36. Minimum numbers of galls (17) were recorded in FD-49-62 followed by FD-1-3 (18), SH-339 (18.667), SH-704 (19.33) and SH-332 (21.0).

Effect of *Meloidogyne incognita* infection on egg mass development:

Data regarding egg mass production revealed that there is significant difference in nematode egg mass production on the roots of each cultivar tested against root knot nematode (*M. incognita*) infection. Statistically ($P > 0.05$) maximum numbers of egg masses (77.0) were produced of FD-19-2 followed by SH-692 (75.6), FD-8-1 (73.3) and SH-5 (68.66). FD-1-3 exhibit minimum number of egg masses (23.3) as compared to all other cultivars. FD-75-3 had number of galls (24.6) followed by FD-49-62, (25.66), FD-51-5 (26.0) and FD-53-1 (29.0).

Effect of *Meloidogyne incognita* infection on nematode population per root system: Nematode population per root system recorded after harvesting revealed that statistically ($P < 0.05$) maximum numbers of nematodes per root system (2607.8) were recorded in FD-19-2 followed by FD-69-1 (2317.5), FD-8-1 (2310) and SH-692 (2250.1). All other cultivars had lower number of nematodes as compared to these cultivars.

Table 1. Effect of *Meloidogyne incognita* infection on plant growth parameters.

Cultivar	No. of Leaves	Tuber weight	Root Weight	Shoot Weight
Cardinal	66.565 ± 1.15 B	45.00 ± 0.82 D	8.92 ± 0.58 NOP	26.97 ± 0.44 E
FD-1-3	66.872 ± 0.33 B	62.33 ± 1.47 B	8.52 ± 1.32 OPQ	29.47 ± 0.44 B
FD-19-2	33.249 ± 0.37 T	17.00 ± 0.82 T	13.78 ± 0.58 AB	17.91 ± 0.37 T
FD-21-10	45.458 ± 0.35 R	24.33 ± 0.78 R	12.87 ± 0.73 BC	19.63 ± 0.44 PQ
FD-35-1	62.366 ± 0.37 DEFG	36.67 ± 0.82 H	9.51 ± 0.58 MN	25.92 ± 0.61 G
FD-35-25	54.238 ± 0.35 MNO	32.33 ± 0.78 M	10.45 ± 0.73 IJKL	22.16 ± 0.53 K
FD-35-36	49.14 ± 0.33 P	29.00 ± 1.47 NO	11.70 ± 1.32 EF	21.21 ± 0.53 LM
FD-49-28	62.147 ± 0.37 EFG	35.33 ± 0.82 IJ	9.48 ± 0.58 MN	25.30 ± 0.37 H
FD-49-62	75.912 ± 0.35 A	84.67 ± 0.78 A	7.97 ± 0.73 Q	29.87 ± 0.53 A
FD-51-5	47.057 ± 0.35 QR	26.67 ± 0.78 PQ	11.81 ± 0.73 DEF	20.90 ± 0.37 N
FD-5-2	48.0450.70 PQ	27.00 ± 0.82 P	11.62 ± 0.58 FG	21.10 ± 0.44 MN
FD-52-7	62.9 ± 1.93 CDEF	36.00 ± 1.47 HI	9.70 ± 1.32 KLMN	25.87 ± 0.44 G
FD-53-1	64.113 ± 1.42 CD	42.00 ± 0.82 EF	9.42 ± 0.58 MNO	26.46 ± 0.53 F
FD-53-2	48.92 ± 0.71 P	28.67 ± 0.78 O	11.45 ± 0.73 FGH	21.20 ± 0.61 LM
FD-53-6	57.344 ± 1.05 JK	33.33 ± 1.47 KLM	10.30 ± 1.32 IJKLM	23.58 ± 0.61 I
FD-69-1	46.495 ± 0.50 QR	25.67 ± 0.82 Q	12.68 ± 0.58 CD	19.86 ± 0.53 P
FD-69-2	60.92 ± 0.77 GH	34.00 ± 1.47 K	9.90 ± 1.32 JKLM	25.22 ± 0.53 H
FD-70-1	55.388 ± 1.27 LMN	32.67 ± 0.78 LM	10.54 ± 0.73 HIJK	22.18 ± 0.44 K
FD-75-3	63.325 ± 0.91 CDE	41.33 ± 0.78 F	9.47 ± 0.73 MN	26.44 ± 0.44 F
FD-76-35	63.555 ± 0.60 CDE	40.00 ± 1.47 G	9.65 ± 1.32 KLMN	26.22 ± 0.37 F
FD-8-1	30.194 ± 0.96 U	20.33 ± 1.47 S	13.81 ± 1.32 A	18.19 ± 0.44 S
FD-8-3	53.88 ± 1.32 NO	30.00 ± 1.47 N	10.77 ± 1.32 GHIJ	22.13 ± 0.44 K
FSD-White	57.457 ± 0.60 JK	33.33 ± 0.78 KLM	9.91 ± 0.73 JKLM	25.11 ± 0.37 H
N-2002-1	63.597 ± 1.01 CDE	36.33 ± 0.78 HI	9.59 ± 0.73 LMN	25.88 ± 0.53 G
N-22	61.406 ± 0.50 FG	34.33 ± 0.78 JK	9.70 ± 0.73 KLMN	25.24 ± 0.61 H
N-30	55.912 ± 0.56 KLM	32.67 ± 0.82 LM	10.51 ± 0.58 IJK	23.20 ± 0.61 J

Cont...

Cultivar	No. of Leaves	Tuber weight	Root Weight	Shoot Weight
N-9619	58.292 ± 0.84 IJ	33.33 ± 0.82 KLM	10.22 ± 0.58 IJKLM	25.12 ± 0.53 H
SH-216	47.034 ± 0.87 QR	26.33 ± 1.47 PQ	12.54 ± 1.32 CDE	20.30 ± 0.61 O
SH-278	56.15 ± 0.52 KL	32.67 ± 1.47 LM	10.77 ± 1.32 GHIJ	23.48 ± 0.37 I
SH-332	66.96 ± 0.81 B	46.67 ± 0.78 C	8.47 ± 0.73 PQ	27.75 ± 0.61 D
SH-339	67.64 ± 1.35 B	61.67 ± 0.82 B	8.42 ± 0.58 PQ	28.14 ± 0.37 C
SH-479	53.014 ± 0.91 O	29.67 ± 0.82 NO	10.96 ± 0.58 FGHI	21.47 ± 0.37 L
SH-5	43.19 ± 1.00 S	23.33 ± 1.47 R	12.84 ± 1.32 C	19.55 ± 0.37 Q
SH-692	31.298 ± 0.49 U	20.33 ± 0.78 S	13.44 ± 0.73 ABC	18.61 ± 0.53 R
SH-701	59.545 ± 0.74 HI	33.67 ± 0.82 KL	9.72 ± 0.58 KLMN	25.16 ± 0.44 H
SH-704	64.369 ± 1.37 C	43.00 ± 1.47 E	9.41 ± 1.32 MNO	26.49 ± 0.61 F
LSD	3.0465	1.0648	0.9176	0.2780

Means sharing same letters are not significantly different from each other at P = 0.05.

Table 2. Effect of *Meloidogyne incognita* infection on nematode reproduction parameters.

Cultivar	Number of Galls	Egg Masses	Nematode Population/ Root System	Reproduction Factor**
Cardinal	21.00 ± 0.84 T	29.00 ± 1.00 Z	2230.60 ± 11.35 CD	1.74 ± 0.01 a
FD-1-3	19.33 ± 0.59 U	24.67 ± 0.73 b	2250.10 ± 20.46 C	1.58 ± 0.01 b
FD-19-2	43.00 ± 0.43 B	77.00 ± 0.59 A	726.80 ± 10.14 U	5.38 ± 0.01 A
FD-21-10	41.33 ± 0.38 C	67.67 ± 0.77 E	736.30 ± 17.76 U	4.17 ± 0.02 E
FD-35-1	29.67 ± 0.84 N	35.00 ± 1.00 VW	1884.80 ± 21.34 G	1.92 ± 0.01 VW*
FD-35-25	39.00 ± 0.55 G	51.67 ± 0.95 N	1010.80 ± 18.80 NO	3.21 ± 0.03 L
FD-35-36	39.67 ± 0.59 EF	57.33 ± 0.73 K	896.30 ± 21.36 S	3.88 ± 0.02 J
FD-49-28	30.33 ± 0.43 M	37.00 ± 0.59 T	1994.50 ± 10.14 F	2.01 ± 0.01 T
FD-49-62	17.00 ± 0.38 X	23.33 ± 0.77 c	2607.80 ± 12.91 A	1.48 ± 0.03 c
FD-51-5	40.00 ± 0.55 E	62.33 ± 0.95 H	883.80 ± 17.02 S	4.00 ± 0.02 H
FD-5-2	40.00 ± 0.56 E	61.67 ± 0.71 I	896.80 ± 11.35 S	3.98 ± 0.01 HI
FD-52-7	28.00 ± 0.59 O	36.33 ± 0.73 U	1554.70 ± 20.46 I	1.95 ± 0.01 U
FD-53-1	24.00 ± 0.43 R	30.67 ± 0.59 Y	2069.50 ± 12.91 E	1.86 ± 0.02 Y

Cont...

Cultivar	Number of Galls	Egg Masses	Nematode Population/ Root System	Reproduction Factor**
FD-53-2	40.00 ± 0.38 E	58.33 ± 0.77 J	939.80 ± 25.34 R	3.97 ± 0.02 I
FD-53-6	36.00 ± 0.83 I	46.00 ± 0.96 P	1011.30 ± 27.30 N	2.25 ± 0.01 P
FD-69-1	41.00 ± 0.56 CD	65.00 ± 0.71 F	830.30 ± 12.91 T	4.13 ± 0.02 F
FD-69-2	31.33 ± 0.59 L	38.67 ± 0.73 S	1228.10 ± 21.36 K	2.11 ± 0.02 R
FD-70-1	39.00 ± 0.43 G	48.33 ± 0.82 O	998.10 ± 17.76 NOP	2.70 ± 0.02 M
FD-75-3	27.67 ± 0.38 O	34.00 ± 0.77 X	2061.50 ± 17.76 E	1.90 ± 0.02 X
FD-76-35	26.67 ± 0.83 P	34.67 ± 0.96 W	1849.00 ± 19.81 H	1.91 ± 0.01 WX
FD-8-1	44.00 ± 0.54 A	75.67 ± 0.68 B	697.80 ± 20.46 V	4.82 ± 0.01 B
FD-8-3	39.33 ± 0.59 FG	52.67 ± 0.73 M	991.30 ± 20.46 OP	3.27 ± 0.01 K
FSD-White	33.00 ± 0.43 J	42.00 ± 0.82 Q	1181.30 ± 17.02 L	2.16 ± 0.02 Q
N-2002-1	25.00 ± 0.38 Q	35.33 ± 0.77 V	1871.10 ± 18.80 G	1.94 ± 0.03 UV
N-22	31.00 ± 0.84 L	38.33 ± 1.23 S	1536.30 ± 25.34 I	2.04 ± 0.02 S
N-30	38.00 ± 0.56 H	46.33 ± 0.71 P	1004.30 ± 21.34 NO	2.60 ± 0.01 N
N-9619	32.00 ± 0.61 K	41.67 ± 0.76 Q	1056.30 ± 12.91 M	2.13 ± 0.02 R
SH-216	40.67 ± 0.42 D	64.00 ± 0.55 G	881.30 ± 27.30 S	4.08 ± 0.01 G
SH-278	38.00 ± 0.37 H	46.00 ± 0.51 P	981.80 ± 19.81 PQ	2.47 ± 0.01 O
SH-332	18.67 ± 0.84 V	26.00 ± 1.23 a	2310.00 ± 25.34 B	1.59 ± 0.02 b
SH-339	18.00 ± 0.56 W	25.67 ± 0.71 a	2317.50 ± 10.14 B	1.58 ± 0.01 b
SH-479	39.33 ± 0.61 FG	54.00 ± 0.76 L	969.30 ± 10.14 Q	3.86 ± 0.01 J
SH-5	41.33 ± 0.42 C	68.67 ± 0.55 D	739.30 ± 19.81 U	4.29 ± 0.01 D
SH-692	43.33 ± 0.38 B	73.33 ± 0.77 C	736.30 ± 18.80 U	4.67 ± 0.03 C
SH-701	32.00 ± 0.84 K	40.67 ± 1.00 R	1270.00 ± 11.35 J	2.12 ± 0.01 R
SH-704	22.67 ± 0.54 S	30.33 ± 0.68 Y	2220.80 ± 27.30 D	1.83 ± 0.01 Z
LSD	0.5344	0.5724	19.608	0.0201

Means sharing same letters are not significantly different from each other at P = 0.05

Minimum number of nematodes were recorded in FD-49-62 (697.8) followed by FD-1-3 (726.8), FD-51-5 & SH-332 (736.3) and SH-339 (739.3).

Effect of *Meloidogyne incognita* infection on nematode reproduction factor: Data regarding nematode reproduction factor on each cultivar (Table 2) revealed that there is significant difference between nematode reproduction factors on each potato cultivar. FD-49-62 exhibit high nematode reproduction factor followed by FD-1-3, SH-339 and SH-332. Magnitude of nematode reproduction factor (5.83) was high FD-19-2 followed by FD-8-1 (4.81), SH-692 (4.67) and SH-5 (4.29).

Root knot nematodes are the most damaging group of plant parasitic nematodes associated with vegetable crops occurring throughout the world. The areas having sandy loam and loam soil type has more root knot nematode (*M. incognita*). Soil type is an important factor affecting nematode movement, growth and reproduction rate and host suitability (Starr *et al.*, 1993; Ogbuji, 2004; Anwar *et al.*, 2007).

Root knot nematode (*M. incognita*) the hidden enemies are causing significant yield losses by attacking on the root systems of the susceptible host plant. Primary symptom of *Meloidogyne incognita* infection is the formation of root galls on the roots of susceptible host plants. Flow of water and nutrients into the plants is greatly influenced by nematode feeding that leads to the reduced yield of the agricultural crops (Roberts, 1987; Sikora and Fernandez, 1990). In addition to nematode infestations, plants with damaged roots become susceptible to other disease causing agents and stress factors including heat, water and nutritional deficiencies (Powell, 1971; Anwar and Din, 1986; Khan *et al.*, 2007). Genotypic/ Varietal response of thirty six different potato cultivars to root knot nematode (*M. incognita*) infection evaluated on the basis of plant growth parameters (number of leaves per plant, average tuber weight of each cultivar, root weight per plant and shoot weight per plant) and nematode reproduction parameters (root galls and egg mass indices, number of nematodes per root system, number of nematodes per 100 ml of soil and nematode reproduction parameter was) was considerably variable among cultivars.

There was not a single cultivar immune to root knot nematode (*M. incognita*) infection. Susceptibility of different vegetable crops and potato cultivars had already been reported by different scientists (Khan *et al.*, 2006; Abbas *et al.*, 2008; Kamran, 2008; Sahi, 2008;

Ubaid, 2009 and Hayat *et al.*, 2012). Results of the present studies were different from Shahzad *et al.*, 1999 and Darban *et al.*, 2003.

Among different potato cultivars tested against root knot nematode (*M. incognita*) infection, pronounced variation has been observed. The variation of the cultivars to nematode infection might be attributed to their genetic makeup and level of resistance mechanism possessed by a particular cultivar (Barham and Winstead, 1957; Anwar and Mckenry, 2002 and Abad *et al.*, 2003).

Status of the host plant to the nematode infection can be assessed by the number of root galls produced by the root systems of the host plants, nematode reproduction and the final nematode population per gram of root or soil at the time of harvest (Gast *et al.*, 1984; Belair and Benoit, 1996 and Davis *et al.*, 2003). The magnitude of nematode reproduction factor (Pf/Pi = nematode final population over initial population) to determine host status is considered to be more accurate measure of nematode-host relationship (Anwar *et al.*, 2000). Cultivar resistance to plant parasitic nematode infection can also be evaluated on the basis on root galls and egg mass numbers (Hussey and Boerma, 1981).

The reliability among potato cultivars in terms of root galls and egg mass indices propose that it is convenient and better intimation of resistance against root knot nematode (*M. incognita*) (Luzzi *et al.*, 1987). These intimations can only be used for preliminary evaluation of the resistance of cultivars against root knot nematode (*M. incognita*) infection and the concluding estimation must be based on the nematode reproduction (Hussey and Janssen, 2004).

All thirty seven cultivars found susceptible because they had root galls and egg mass indices >3 and rate of reproduction >1. Similar results of root galls, egg mass indices and nematode reproduction rate have also been reported (Hayat, 2011). The amount of eggs laid by female nematodes are influenced by environmental conditions and host status to a particular nematode species or populations (Luc *et al.*, 2005; Ashoub and Amara, 2010).

CONCLUSION

Among different potato cultivars tested against root knot nematode (*M. incognita*) FD-19-2, FD-8-1, SH-292, SH-5 and FD69-1 exhibit high root galls, egg mass indices and nematode reproduction factor and found highly susceptible to root knot nematode infection. Magnitude of nematode reproduction factor, root galls and egg

mass indices were less in FD-49-62, FD-1-3, SH-339, SH-332 cultivars.

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